PROJECT SUMMARY

Title:	**Occurrence of Antibiotics in Wastewater Effluents and Their Mobility in Soils: A Case Study for Wisconsin**
Project I.D.:	DNR Project #169
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Period of Contract:	July 1, 2001 to June 30, 2003

Background/Need:

Recent reports on the detection of pharmaceuticals in water bodies and waste streams in Europe and the United States have raised some environmental concerns. Sulfonamides have been detected in leachate from a Danish landfill, in Berlin drinking water wells for which 80% of the groundwater was bank-filtered surface water, and in groundwater in Germany. Studies in the U.S. have identified antibiotics (sulfonamides and trimethoprim) in groundwater down gradient from a landfill containing hospital waste, in water supply wells in a Nebraska bank filtration site and in groundwater from Washington. There is an increasing interest among scientists, policy makers and industry personnel in the U. S. to survey the nation's water resources for human and veterinary pharmaceuticals, steroidal hormones, etc. The United States Geological Survey (USGS) is leading the efforts by collecting stream samples from several locations across the country to obtain baseline information regarding these emerging organic contaminants. Their efforts need to be augmented at the local level by systematically screening potential sources (e.g., wastewater treatment effluent, on-site wastewater systems, animal waste lagoons) to obtain a better understanding of the transport pathways and environmental fate of pharmaceuticals. Compared to conventional organic contaminants (e.g., pesticides, PAHs, PCBs), little information is available on the environmental fate and transport of antibiotics. The molecular properties of the antibiotics would favor sorption to soil components and interaction with metal ions in soils. However, our ability to predict mobility and fate of antibiotics in surface and subsurface systems is hampered by a lack of understanding of the fundamental processes governing their environmental reactivity.

Objectives:

i) Conduct a screening for five different classes of antibiotics, namely, sulfonamides, macrolides, tetracyclines, fluoroquinolones, and beta-lactams, in the effluents from wastewater treatment systems, and in adjacent groundwater monitoring wells, and

ii) Determine the environmental fate and transformation of selected antibiotics, namely, tetracyclines and fluoroquinolones.

Methods:

Our sampling activities spanned a range of wastewater treatment methods, community size served by the facility, receiving water body impacted by effluent discharge and geographical locations within the state. Samples were collected from seven different wastewater treatment facilities (Green Bay, Oshkosh, Lake Geneva, Barron-Cameron, Hayward, Spooner, Middle River Health Care), adjacent groundwater monitoring wells at two sites (Lake Geneva, Spooner), and two on-site wastewater treatment systems. In addition, three water supply wells in La Crosse under the influence of surface water were sampled. Filtered water/wastewater samples collected from the study sites were overnight on ice (4 ^{o}C) to the USGS, Ocala, FL for antibiotic analysis. The samples were extracted following established procedures and analyzed using a LC/MS fitted with atmospheric pressure ionization (LC/API-MS) interfaces. Dr. Michael

T. Meyer (USGS) who has developed a method to extract and analyze 25 antibiotic compounds in water/wastewater/manure extracts, provided analytical and interpretive support for this study.

Batch experiments were used to quantify the sorption of tetracycline and fluoroquinolone to well-known soil minerals (hydrous oxides of Fe and Al). These experiments involved reacting $Al(OH)_3$/$Fe(OH)_3$ to the selected antibiotics (tetracycline and ciprofloxacin) in centrifuge tubes. After equilibration at 25 ^oC at 7 rpm for 24 h, the suspensions were centrifuged and filtered (0.2 μm). Antibiotic concentrations (HPLC, by UV detection), $^3H/^{14}C$ activities (liquid scintillation counting (LSC)), and pH were determined in the supernatant. LSC provided a measure of the total activity contributed by these compounds, while HPLC allowed us to determine whether significant degradation occurred during the batch experiments. The amount of antibiotics sorbed was calculated from the difference between solution-phase $^{14}C/^3H$ activities in equilibrated blanks and suspensions (corrected for any degradation).

Results and Discussion:

A total of eight antibiotic compounds were detected (1-5 compounds per site), including three sulfonamides (sulfamethazine, sulfamethoxazole, and sulfadimethoxine), two tetracyclines (tetracycline, chlortetracycline), one fluoroquinolone (ciprofloxacin), one macrolide (erythromycin) and trimethoprim. Detected antibiotics fall into the following classes for the frequency of detection: sulfonamides > tetracyclines > fluoroquinolones > trimethoprim > macrolides. The USGS nationwide stream survey lists the above compounds among the 30 most frequently detected organic contaminants and the levels detected by us are in agreement with those found in the literature.

Contrasting sorption behavior of tetracycline and ciprofloxacin to Al and Fe hydroxides was observed. Compared to tetracycline the following trends were observed for ciprofloxacin: (i) strong pH-dependence (ii) minor differences between LSC and HPLC measurements after both 1 day and 1 week, and (iii) higher removal level - maximum sorption of ≈ 72%, unaffected by reaction time, around neutral pH. Tetracycline sorption exhibits mild pH-dependence and a significant difference existed between LSC- and HPLC-determined removal levels, highlighting the importance of transformation reactions. Our efforts to identify degradation products did not reveal the presence of hydrolyzed forms (most plausible reaction products) or other compounds. However, elevated soluble Al levels observed in the presence of tetracycline indicated that ligand-promoted dissolution of $Al(OH)_3$ could be occurring. The presence of Al-tetracycline complexes in solution helps explain the difference between HPLC and LSC quantifications. Our research findings will increase understanding of the environmental occurrence, fate and transformation characteristics of these emerging organic contaminants.

Conclusions/Implications/Recommendations:

Antibiotics were detected in wastewater influent/effluent, adjacent groundwater monitoring wells and in samples from on-site wastewater treatment systems. However, the soluble levels were extremely low (< 10 μg/L), and importantly were unaffected by the size of the wastewater treatment facility. Future monitoring programs can be limited to the eight compounds detected in our study.

Key Words:

Antibiotics, wastewater treatment, sulfonamide, tetracycline, fluoroquinolone, macrolide, trimethoprim, ciprofloxacin, sorption, Al hydroxide, Fe hydroxide, transformation

Funding:

Wisconsin Department of Natural Resources
Wisconsin Department of Agriculture, Trade, and Consumer Protection

Final Report:

A final report containing more detailed information on this project is available for loan from Wisconsin's Water Library, University of Wisconsin - Madison, 1975 Willow Drive, Madison, Wisconsin 53706 (608) 262-3069.